

**Adults and Metacercariae of Three Microphallid Trematodes,  
Including a New Species of the Genus *Maritrema*,  
from Queensland, Australia**

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**Abstract**

Encysted metacercariae of three microphallid trematodes were found in freshwater prawns, *Caridina nilotica* (Atyidae) and *Macrobrachium australiense* (Palaemonidae), and a freshwater crayfish, *Cherax dispar* (Parastacidae), from the Brisbane River, Queensland, Australia. Their adults were obtained from chicks and mice in feeding experiments. *Maritrema brevisacciferum* sp. n. (= *M. oocysta* of Deblock and Pearson, 1968) is described and figured on the basis of metacercariae from *C. nilotica* and adults from chicks. It is compared with related known species. Metacercariae and adults are described and figured of *Microphallus minutus* Johnston, 1948, from *C. dispar* and mice, and of *Pseudolevinseniella anenteron* Deblock and Pearson, 1968, from *M. australiense* and mice, respectively.

**Key words:** Microphallids, a new species, life cycles, trematodes, Australia

Many species of microphallid trematodes have been reported from mammals and birds in Australia, and the life cycles of some of them have been elucidated (Smith, 1974, 1981, 1983). Microphallid metacercariae of three species were found in prawns and a crayfish in Queensland, Australia. They were experimentally raised to adults in chicks and mice. One of the three has proved to represent a new species of the genus *Maritrema* Nicoll, 1907, and the two others have been identified as known species. This paper describes the metacercariae and adults of the three species.

**Materials and Methods**

Freshwater prawns, *Caridina nilotica* (Atyidae) and *Macrobrachium australiense* (Palaemonidae), and a freshwater crayfish, *Cherax dispar* (Parastacidae), were collected in the Brisbane River at Twin Bridges near Fernvale,

Queensland, Australia, from November 1988 to February 1989. They were examined fresh for metacercariae. Metacercariae isolated from them were fed to young mice and 1-day-old chicks with a stomach tube. The animals were examined for adult flukes at irregular intervals from 1 to 5 days PI.

Metacercarial and adult worms obtained were flattened, fixed in AFA, stained with either Heidenhain's iron hematoxylin or alum carmine and mounted in Canada balsam. All measurements (length by width) are given in micrometers unless stated otherwise. The specimens are deposited in the collections of the Queensland Museum (QM), Brisbane, and the National Science Museum, Tokyo (NSMT).

Family Microphallidae Ward, 1901  
Subfamily Maritreminae Nicoll, 1909  
Genus *Maritrema* Nicoll, 1907  
*Maritrema brevisacciferum* sp. n.  
(Figs. 1–5)

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*Maritrema oocysta* (Lebour, 1907) Rothschild, 1942, *sensu* Deblock and Pearson, 1968, p. 459.

Encysted metacercariae were found in various

organs (muscles, gills, hepatopancreas, *etc.*) of the cephalothorax of the prawn, *C. nilotica*. Worms were usually folded in their cysts. They spontaneously excysted easily, when they were kept in 0.6% saline after having been taken out of their hosts.

Many metacercariae were fed to 13 chicks, which were examined 2 to 5 days PI, and to 1 mouse, which was examined 2 days PI. Adult worms were recovered from the small intestine of all the chicks, but no worms were found in the mouse.

*Metacercaria*. Cysts (Fig. 1) oblong, 0.15–0.23 by 0.12–0.14 mm; cyst wall colorless, hyaline, 0.01–0.02 mm thick, consisting of 3 layers; innermost layer opaque in part.

Ten whole-mounts measured (NSMT-P1 3671; Fig. 2). Resembling adult worms in general morphology. Body 232–288 by 144–168; forebody 120–160 long, 52–57% of total body length. Oral sucker 24–27 by 30–32. Pharynx 17–19 by 11–13. Esophagus 19–29 long; intestinal ceca 56–64 long. Ventral sucker 33–40 by 40–43; sucker with ratio 1: 1.25–1.37. Testes 33–40 by 32–36. Cirrus pouch 96–104 by 24–29; ratio of length of cirrus pouch to width of ventral sucker 1: 2.22–2.60. Ovary 32 by 29–40. Excretory vesicle lined with large cells; flame cell formula  $2 [(2+2) + (2+2)] = 16$ .

*Adult*. Ten 5-day-old whole-mounts measured (Figs. 3–5). Body oval to lanceolate-oblong, widest about testicular level, spinose, 264–416 by 168–200; forebody 144–208 long, 48–54% of total body length. Oral sucker 25–29 by 32–33. Prepharynx short. Pharynx oblong, 17–22 by 14–19. Esophagus slender, 17–40 long; intestinal ceca short, assuming an inverted Y-shape together with esophagus, not extending to ventral sucker, 64–88 long. Ventral sucker postbifurcal, 26–51 by 40–49; sucker width ratio 1: 1.25–1.55.

Testes globular to elliptical, symmetrical,

lateral, straddling boundary between middle and posterior thirds of body, 48–72 by 40–52. Cirrus pouch muscular, thick-walled, clavate, bending posteriorly at distalmost part, lying almost transversely between intestinal ceca and ventral sucker, 88–112 by 27–33; ratio of length of cirrus pouch to width of ventral sucker 1: 2.04–2.59; seminal vesicle internal, elongate-clavate, occupying proximal one-third of cirrus pouch; pars prostatica small, accompanied by internal prostatic cells; cirrus aspinose, filiform, long, protrusible. Genital atrium simple, fairly large. Genital pore sinistral or anterosinistral to ventral sucker. Ovary globular, located between ventral sucker and right testis, 40–72 by 30–40. Middle part of oviduct swollen, coiled once, storing a small amount of sperm, with a posteriorly directed Laurer's canal; true seminal receptacle absent. Ootype complex usually posterior to ventral sucker. Uterus first running anteriorly beyond ventral sucker and then turning backward into hindbody, much coiled there, extending nearly to intestinal cecum on either side of body, serving as a uterine seminal receptacle; metraterm well developed, not very long, with a well-developed sphincter near its opening into genital atrium. Eggs operculate, embryonated, 16–19 by 8–11. Vitelline follicles standing in a line lying along body margin, bending inward and passing along anterior border of testis on either side of hindbody. Excretory vesicle V-shaped, each arm extending to midlevel of testes.

Type final host: Chick (experimental).

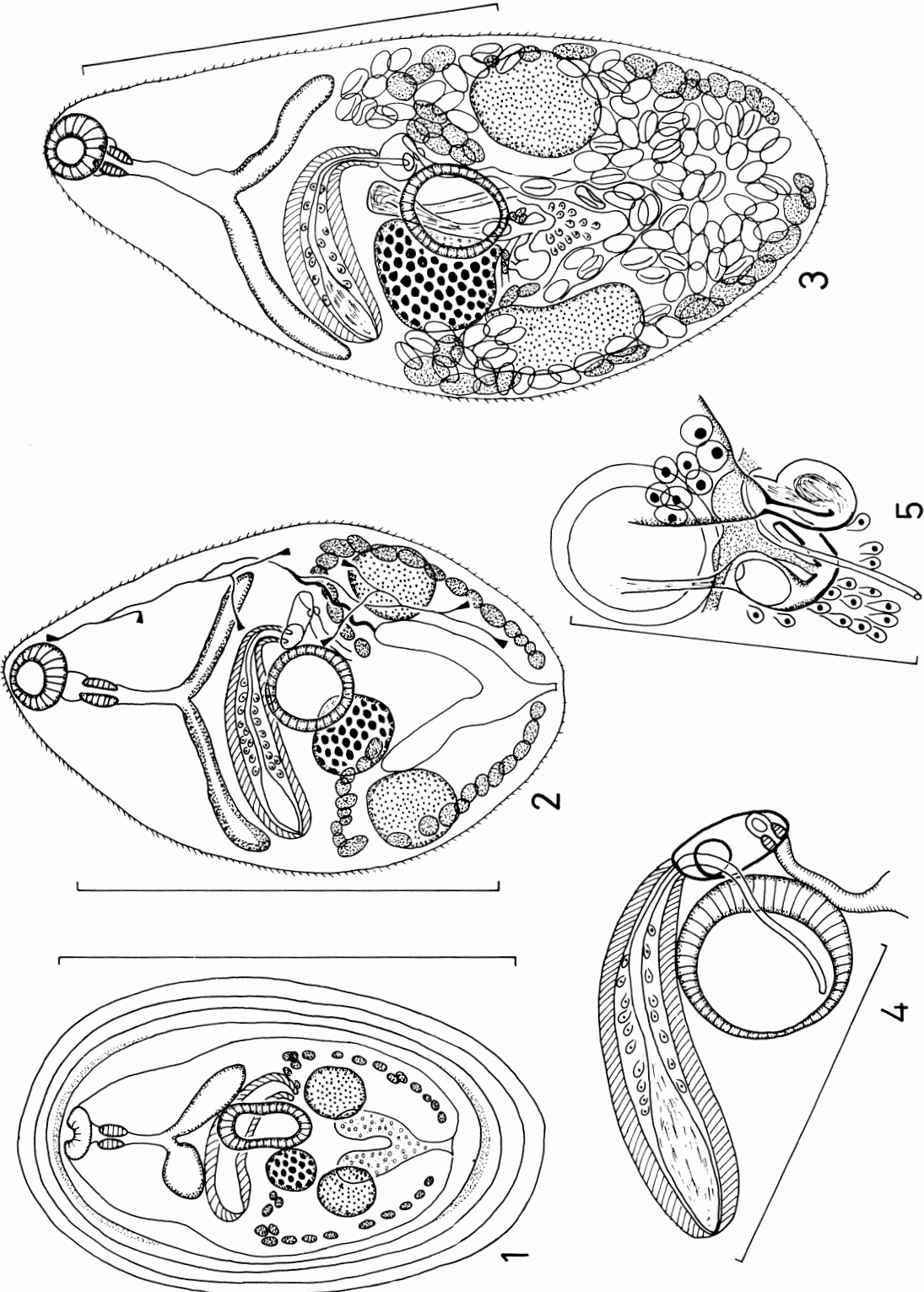
Site of infection: Small intestine.

Specimens: Holotype, QM GL 13087 (5-day-old adult); paratypes, QM GL 13088 (seven 5-day-old adults) and NSMT-P1 3668–3670 (fourteen 2-, 3- and 5-day-old adults).

*Discussion*. This new freshwater species, *Maritrema brevisacciferum* sp. n., is identical to the trematode that Deblock and Pearson (1968)

Figs. 1–5 *Maritrema brevisacciferum* sp. n. 1: Encysted metacercaria found in the prawn, *Caridina nilotica*, from the Brisbane River, Queensland, Australia, formalin-fixed, semi-schematic. 2: Excysted metacercarial worm, excretory system being semi-schematically drawn, ventral view. 3: Adult worm experimentally obtained from a chick 5 days PI, holotype, ventral view. 4: Terminal genitalia, adult worm, paratype, ventral view. 5: Ootype complex, adult worm, paratype, dorsal view.

(Scale bars: 0.2 mm in Figs. 1–3; 0.1 mm in Figs. 4 and 5.)



recorded as *M. oocysta* (Lebour, 1907) Rothschild, 1942, from the digestive tract of a water rat, *Hydromys chrysogaster* (Rodentia: Muridae), from Moggill, and a cormorant, *Anhinga novaehollandiae* (Pelecaniformes: Anhingidae), and a grebe, *Podiceps ruficollis* (Podicipediformes: Podicipedidae), from Kolo, all in Queensland, Australia. We have confirmed this by a direct comparison between the present adults and several of the adults obtained from the water rat. The trematode that Obendorf and Smales (1985) and Smales *et al.* (1990) recorded as *M. oocysta* from water rats in Tasmania, and Victoria and South Australia, respectively, could also be the new species.

The new species certainly resembles *M. oocysta* (= *M. humile* Nicoll, 1907 = *Pseudomaritrema innae* Leonov, 1958) in Europe (Deblock, 1971, 1975; Deblock and Capron, 1960) particularly in the thick-walled muscular cirrus pouch and the aspinose filiform cirrus. However, it differs from the latter in that it occurs in a freshwater, instead of brackish-water, habitat; the metacercaria encysts in the prawn instead of its sporocyst still within the brackish-water snail host; the oral sucker is distinctly smaller than, instead of practically equal to, the ventral sucker; the cirrus pouch is smaller, the ratio of its length to the width of the ventral sucker being 1: 2.04–2.60 instead of 1: more than 3.5 (calculated from Deblock's descriptions).

There seem to be some similarities between the new species and *M. ornithorhynchi* Hickman, 1955, in morphology and habitat. The latter lives in the platypus *Ornithorhynchus anatinus* (Monotremata: Ornithorhynchidae) of Tasmania (Hickman, 1955). Its type specimens have not been available to us for reexamination, but we were able to examine three mounted adult specimens which Dr. Hickman kindly sent to one of us (J. P.). In the adult morphology, the present new species is readily differentiated from *M.*

*ornithorhynchi* (our observation) by a smaller body, a larger ventral sucker (the sucker width ratio being 1: 1.25–1.55 instead of 1: 1.12–1.25), a smaller cirrus pouch (its length being 2.04–2.59, instead of 3.2–3.5, times the width of the ventral sucker), a smaller seminal vesicle occupying the proximal one-third, instead of more than two-thirds, of the cirrus pouch. The new species has an aspinose filiform cirrus and no seminal receptacle. It is said that in *M. ornithorhynchi*, the ejaculatory duct opens on a small papilla in the genital atrium and the seminal receptacle is present (Hickman, 1955). We were unable to confirm these features definitely in the three specimens of the species because they were poorly stained and the region of the ootype complex was obscured by the uterus occupying all available spaces in the hindbody.

The new species appears to be similar to *M. calvertense* Smith, 1974, from a duck, *Anas castanea* (Anseriformes: Anatidae), and plovers, *Charadrius cucullatus* and *C. melanops* (Charadriiformes: Charadriidae) of Tasmania. However, the latter is a brackish-water species and has a much larger cirrus pouch (Smith, 1974, 1981).

#### Subfamily Microphallinae Ward, 1901

##### Genus *Microphallus* Ward, 1901

##### *Microphallus minutus* Johnston, 1948

(Figs. 6–10)

*Microphallus minutus* Johnston, 1948, pp. 93–97, figs. 1–8.

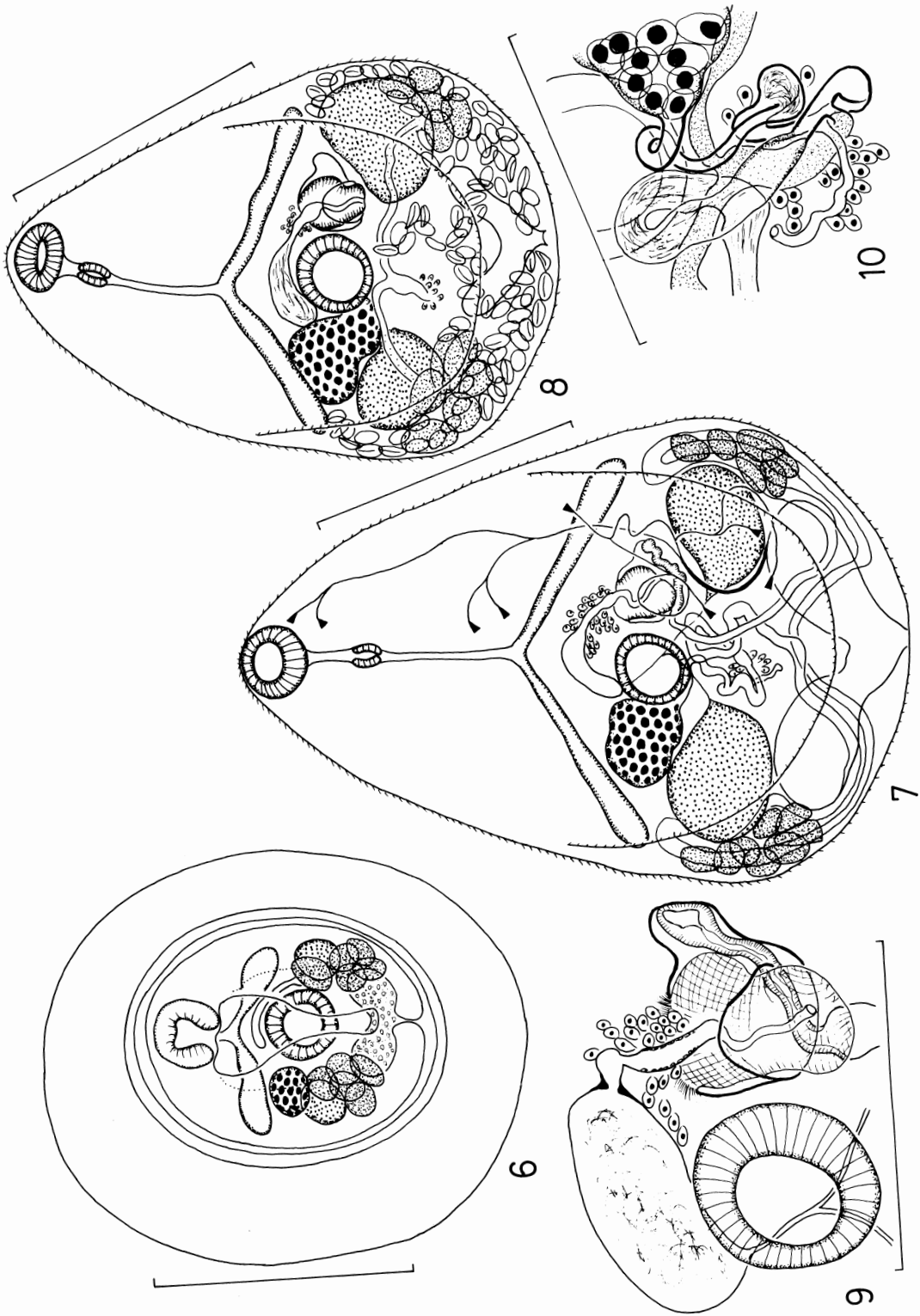
*Microphallus minutus*: Deblock and Pearson, 1969, pp. 394–395, figs. 4–6.

Encysted metacercariae were found in the muscles of the abdomen of the crayfish, *Cherax dispar*. Worms were usually bent double in their cysts.

Many metacercariae were fed to two mice and two chicks, which were examined 2 days PI. Eight

Figs. 6–10 *Microphallus minutus* Johnston, 1948. 6: Encysted metacercaria found in the crayfish, *Cherax dispar*, from the Brisbane River, Queensland, Australia, formalin-fixed, semi-schematic. 7: Excysted metacercarial worm, excretory system being semi-schematically drawn, ventral view. 8: Adult worm experimentally obtained from a mouse 2 days PI, ventral view. 9: Terminal genitalia, adult worm, ventral view. 10: Ootype complex, adult worm, dorsal view.

(Scale bars: 0.2 mm in Figs. 6–8; 0.1 mm in Figs. 9 and 10.)



adult worms were recovered only from the small intestine of one of the two mice.

**Metacercaria.** Cysts (Fig. 6) elliptical, 0.31–0.54 by 0.29–0.54 mm; cyst wall consisting of 2 layers; outer layer white, opaque, sticky, thick, 0.07–0.32 mm thick; inner layer colorless, hyaline, thin, 0.23–0.26 by 0.19–0.24 mm, about 0.01 mm thick.

Worms (NSMT-PI 3672 and 3673; Fig. 7) closely resembling adult worms in general morphology and size except for a much smaller seminal vesicle. Body margins infolded ventrally. Excretory vesicle V-shaped, lined with large cells; flame cell formula  $2 [(2 + 2) + (2 + 2)] = 16$ .

**Adult.** Eight 2-day-old whole-mounts (NSMT-PI 3674) measured (Figs. 8–10). Body 376–408 by 256–312; forebody 216–240 long, 57–59% of total body length. Oral sucker 32–40 by 51–56. Pharynx 22–24 by 16–17, with five anterior protuberances. Esophagus 48–80 long; intestinal ceca 128–152 long. Ventral sucker 51–56 in diameter; sucker width ratio 1: 1.00–1.06. Testes almost globular, 72–96 by 40–64. Seminal vesicle 56–80 by 16–40. Male copulatory organ or papilla cone-like, straight, 45–67 by 40. Genital atrium simple. Ovary 56–72 by 48–80. Oviduct long; its middle part swollen, sometimes coiled once, storing a small amount of sperm. Seminal receptacle not seen. Uterine seminal receptacle present. Laurer's canal present. Eggs 16–20 by 10–11; contents not worked out. Vitelline follicles about six on each side of body.

**Discussion.** This freshwater species *Microphallus minutus* was first described by Johnston (1948) on the basis of the adult stage found in the small intestine of the water rat *Hydromys chrysogaster* from the Murray River at Tailem Bend, South Australia. Later Deblock and Pearson (1969) briefly described their own adult worms found in water rats from Moggill

and Palmwood, Queensland. Obendorf and Smales (1985) and Smales *et al.* (1990) recorded the species from water rats in Tasmania, and Victoria and South Australia, respectively.

This is the first report of the metacercaria of the species. The morphology of the present experimental adults agrees well with the descriptions by Johnston (1948) and Deblock and Pearson (1969).

Subfamily Pseudolevinseniellinae Deblock, 1971

Genus *Pseudolevinseniella* Tsai, 1955

***Pseudolevinseniella anenteron* Deblock and Pearson, 1968**

(Figs. 11–16)

*Pseudolevinseniella anenteron* Deblock and Pearson, 1968, pp. 459–462, figs. 1–4.

*Allopseudolevinseniella anenteron*: Yamaguti, 1971, p. 601.

Encysted metacercariae were found mainly in the muscles of the pleopods of the prawn, *M. australiense*. Worms were usually extended in their cysts. Some bore incompletely formed eggs in the uterus.

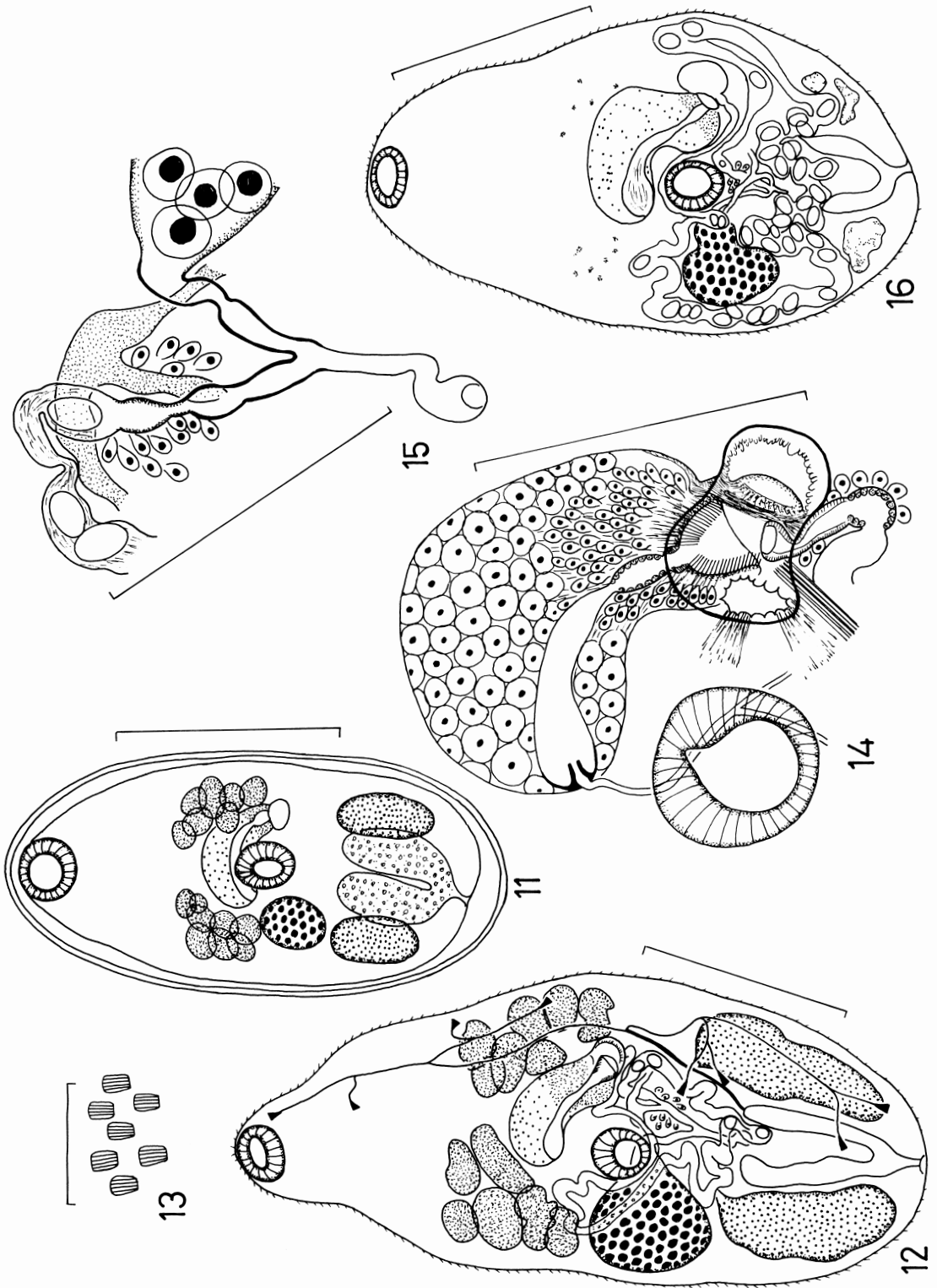
Many isolated metacercariae were fed to six mice, which were examined 2 and 3 days PI, and to one chick, which was examined 2 days PI. Only three adult worms were recovered from the small intestine of two mice 2 days PI. One of them was partly broken during preparation.

**Metacercaria.** Cysts (Fig. 11) oblong, 0.31–0.44 by 0.15–0.22 mm; cyst wall single-layered, transparent, thin, about 0.01 mm thick.

Worms resembling adults; seven whole-mounts (NSMT-PI 3675) measured (Figs. 12–15). Body 528–688 by 256–336; forebody 304–480 long, 60–76% of total body length. Tegumental scales large, with 4–5 longitudinal ridges. Large, dark-stained cells (or possibly parenchymal cells themselves) seen in

Figs. 11–16 *Pseudolevinseniella anenteron* Deblock and Pearson, 1968. 11: Encysted metacercaria found in the prawn, *Macrobrachium australiense*, from the Brisbane River, Queensland, Australia, formalin-fixed, semi-schematic. 12: Excysted metacercarial worm, excretory system being semi-schematically drawn, ventral view. 13: Tegumental scales, metacercarial worm. 14: Terminal genitalia, metacercarial worm, ventral view. 15: Ootype complex, metacercarial worm, dorsal view. 16: Adult worm experimentally obtained from a mouse 2 days PI, ventral view.

(Scale bars: 0.2 mm in Figs. 11–12 and 14; 0.1 mm in Figs. 15 and 16; 0.025 mm in Fig. 13.)



parenchyma. Oral sucker 40–56 by 49–56. Pharynx absent. No digestive tract seen. Ventral sucker 40–56 in diameter; sucker width ratio 1: 0.71–1.00. Testes 120–192 by 48–112. Cirrus pouch 104–160 by 48–80. Ovary 104–136 by 80–112. Vitelline follicles anterolateral to cirrus pouch, about six on each side of body. Excretory vesicle V-shaped, lined with large cells; flame cell formula 2 [(2 + 2) + (2 + 2)] = 16.

*Adult.* Two 2-day-old whole-mounts (NSMT-PI 3676 and 3677) measured (Fig. 16). Body 480–528 by 248–304; forebody 288–320 long, 60% of total body length. Dark-stained cells still seen in parenchyma. Oral sucker 33–48 by 56–59. Ventral sucker 48–51 in diameter; sucker width ratio 1: 0.86. Testes almost completely degenerated. Cirrus pouch 112–144 by 48–64. Seminal vesicle slender. Pars prostatica elliptical, rarely constricted once. Prostatic cells many, large, filling up proximal two-thirds of cirrus pouch. Terminal region of male duct cup-like, lined with spines, opening wide into genital atrium, with many small uninucleate gland cells located compactly in distal one-third of cirrus pouch. Genital atrium large, divided into two (right and left) parts by a crescentic ridge on its bottom, right part receiving male duct and metraterm. Ovary 80–96 by 48–88. Vitelline follicles almost completely degenerated. Eggs 20–24 by 13–17; contents not worked out.

*Discussion.* In morphology, the present metacercaria agrees well with that described by Deblock and Pearson (1968) from the muscles of the prawns, *M. australiense* of the Brisbane River at Moggill and *M. australe* of the McIntyre River at Goondiwindi, Queensland. There has so far been known only the metacercarial stage (Deblock and Pearson, 1968). The natural final host is still unknown. In the present feeding experiments, the testes and vitellaria degenerated within 2 days PI. This suggests that the life span of the adult in the final host is short, possibly two days or a little more. The trematode does not have the intestinal tract, which probably means that the adult lives on food stored as it is a metacercaria and hence is short-lived. It seems likely that the dark-stained cells in the parenchyma store food.

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